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According to the National System of Toxic-Pharmacological Information (Sinitox), accidents involving venomous animals are among the three main causes of human intoxications, along with medication and pesticide poisonings. From 2009 to 2019, there was a 200% increase in the number of bee sting accidents, with a lethality rate of 0.23% of cases, which is close to or even higher than the lethality rates of snake and scorpion envenomations in various regions of Brazil. Many studies aim to better understand the composition of bee venom and the effects of bee envenomation; however, studies addressing the severity of bee envenomation are scarce. Currently, the diagnosis of bee sting accidents is made by counting the stingers on the victim's body, biochemical tests, and the patient's clinical manifestations. However, this approach is time-consuming and presents limitations, as these parameters are not sufficient to determine the extent of envenomation or the amount of circulating venom in the bloodstream. Given the increasing number of bee sting accidents and the limitations in diagnosing patients, this study aimed to develop an analytical approach using mass spectrometry to detect the main toxins of *Apis mellifera* bee venom in biological matrices such as human blood serum and, consequently, to assess the severity of bee sting accidents by comparing experimental results with laboratory-prepared biological matrices. As this study is part of a public-private research and development agreement, the methodological steps cannot be detailed; however, the use of the Selected Reaction Monitoring (SRM) technique with a Q-Exactive (Thermo Fisher) mass spectrometry device is highlighted. A specific toxin from bee venom was monitored in human blood serum biological matrices and bee venom at different concentrations after the enzymatic digestion. As a result, the chosen toxin was detected and quantified in the biological matrices through the precursor ion 462.0923 m/z [M+5H]⁵⁺ and its fragment ions 228.1343 m/z [M+H]⁺ and 299.1714 m/z [M+H]⁺, the precursor ion 432.6108 m/z [M+6H]⁶⁺ and its fragment ions 700.4941 m/z [M+H]⁺, 587.4100 m/z [M+H]⁺, 487.3063 m/z [M+2H]²⁺, 228.1343 m/z [M+H]⁺, 299.1714 m/z [M+H]⁺, 398.2398 m/z [M+H]⁺ and 511.3239 m/z [M+H]⁺, and the precursor ion 756.4634 m/z [M+2H]²⁺ and its fragment ions 1412.8512 m/z [M+H]⁺, 1299.7671 m/z [M+H]⁺, 1198.7194 m/z [M+H]⁺, 706.9262 m/z [M+2H]²⁺, 650.3872 m/z [M+2H]²⁺, 549.3395 m/z [M+2H]²⁺, 464.2867 m/z [M+2H]²⁺, 314.2074 m/z [M+H]⁺, 472.2766 m/z [M+H]⁺ and 490.3130 m/z [M+2H]²⁺. This preliminary study highlights the high potential of the

SRM strategy, which can accurately measure the relative or absolute concentrations of particular molecules such as peptides/proteins in biological matrices. This approach can be useful in integrated studies aimed at developing rapid devices to assess the severity of bee sting accidents.

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